CLAIMS

1. A method for detecting methylated nucleic acids comprising the steps of:

(i) contacting a nucleic acid sample suspected of containing methylated nucleotides with an oligonucleotide probe under suitable conditions for nucleic acid hybridization, said oligonucleotide probe characterized in that,

(a) it comprises a first stem labeled with a fluorophore moiety, a loop sequence having a region of nucleotides complementary to at least a region of the nucleic acid sample, which region of the nucleic acid sample is susceptible to methylation, and a second stem labeled with a quencher moiety that is capable of quenching the fluorophore moiety when in spatial proximity to the fluorophore moiety; and

(b) the nucleotides forming the first stem are capable of moving into spatial proximity with the nucleotides forming the second stem when the probe is dissociated from the nucleic acid sample;

(ii) altering the hybridization conditions such that the oligonucleotide probe dissociates from unmethylated nucleic acids but remains hybridized to methylated nucleic acids; and

(iii) measuring the change in fluorescence;

wherein an increase in fluorescence indicates methylated nucleotides in said nucleic acid sample.

2. A method according to claim 1 wherein the oligonucleotide probe dissociates from the target nucleic acid sample according to step (ii) the first and second stem hybridize together causing quenching of the fluorophore moiety.

3. A method according to claim 1 wherein the loop sequence contains at least 10 nucleotides.

4. A method according to claim 1 wherein the loop sequence contains at least 35 nucleotides.

5. A method according to claim 1 wherein the loop sequence contains at least 25 nucleotides.

6. A method according to claim 1 wherein the loop sequence contains from about 15 to about 20 nucleotides.

7. A method according to claim 1 wherein when the loop sequence is complementary to a portion of a nucleic acid sequence that undergoes methylation when a cell transforms from a normal state to a cancerous state.

8. A method according to claim 1 wherein when the loop sequence is complementary to a portion of a Myf-3 nucleic acid sequence that undergoes methylation when a cell transforms from a normal state to a cancerous state.

9. A method according to claim 8 wherein the loop sequence is complementary to at least one of the sequences selected form the group consisting of:

(i) 5' GCG GCG ACT CCG ACG CGT CCA GCC CGC GCT CC 3' (SEQ ID NO: 1);

(ii) 5' TTA TAC CGC AGG CGG GCG AGC CGC GGG CGC TCG CT 3' (SEQ ID NO: 2); and

(iii) 5' CCG AGA GCC CTG CGG GGC CCG CCC TCC TGC TGG CG 3' (SEQ ID NO: 3).

10. A method according to claim 1 wherein when the loop sequence is complementary to a portiono f a glutathione-S-transferase-II(pi) nucleic acid sequence that undergoes methylation when a cell transforms from a normal state to a cancerous state.

11. A method according to claim 10 wherein the loop sequence is complementary to at least one of the sequences selected from the group consisting of:

(i) 5' CTC CAG CGA AGG CCT CGC GGC CTC CGA GCC TTA TAA G 3' (SEQ ID NO: 4); and

(ii) 5' GGG GAC GCG GGC CGC GCG TAC TCA CTG GTG GCG A 3' (SEQ ID NO: 5).

12. A method according to claim 1 wherein when the loop sequence is complementary to a portion of a calcitonin nucleic acid sequence that undergoes methylation when a cell transforms from a normal state to a cancerous state.

13. A method according to claim 1 wherein the method is used to detect abnormally methylated gene sequences in prostate cancer tissues.

14. A method according to claim 1 wherein the hybridization condition that is altered during the hybridization reaction is the temperature of the hybridization reaction.

15. A method according to claim 1 wherein the stem sequences do not hybridise to the target gene and are of a sufficiently short length to avoid non-specific binding between the stem and any other nucleic acid sequence in the reaction mixture.

16. A method according to claim 1 wherein the stem sequences are at least about 4 to 8 nucleotides in length.

17. A method according to claim 1 wherein at least a cystosine in at least one of the stem sequences contains a methylated cystosine residue.

18. A kit for distinguishing methylated and non-methylated nucleic acid sequences, comprising a labeled oligonucleotide probe, wherein said labeled oligonucleotide probe comprises a fluorophore moiety, a loop sequence, and a quencher moiety, and wherein said loop sequence has a region of nucleotides complementary to at least a region of the nucleic acid sample, which region of the nucleic acid sample is susceptible to methylation.